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Periphyton Productivity and
Radionuclide Accumulation in the
Columbia River, Washington, U.S.A.*

by

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(with 5 figs.)

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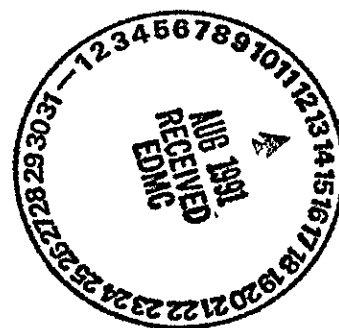
INTRODUCTION

It is generally recognized that the periphyton community comprises the main source of primary production in streams, especially in the smaller, rapidly flowing ones. On the other hand, large rivers such as the Columbia usually contain a significant phytoplankton community, although the components are usually derived from the periphyton and lentic habitats along the river's course.

The autotrophic nature of these organisms and their large surface to volume ratio results in the concentration of certain radionuclides by several orders of magnitude over that of the ambient water. This makes the algae of considerable interest from both a radiobiological and ecological viewpoint. The present study was undertaken as an investigation preliminary to more definitive studies of mineral cycling within the Columbia River. There were two initial objectives: (1) to determine which biological and environmental factors are most influential in governing the amount of ^{32}P and ^{65}Zn accumulated from the river by the periphyton, and (2) to study the relationships between some commonly used indices of production and certain environmental factors. It was also hoped to gain some insight as to the method of sorption of these radionuclides. Data presented are for the period August 1963 to May 1964.

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METHODS AND MATERIALS

Water from the Columbia River is used as a coolant in the reactors at Hanford; during the process the ionic constituents are subjected to intense neutron bombardment which transmutes many of the stable ions to the radioactive state. This water, after further processing, is returned to the river and then contains numerous radioactive isotopes of which more than 60 have been identified (WILSON, ed. 1965).



Fig. 1. Periphyton sampling apparatus.

Twenty-one microscope slides were exposed for two-week periods in the apparatus shown in Fig. 1. The sampler was located in a side channel of the Columbia River below the reactors. The riffle is shown at low water in Fig. 2. Water velocities fluctuate seasonally due to runoff and daily because of regulation by a power dam approximately 48 km upstream. Daily water level fluctuations of two meters are common in the study area. REESE (1937), GUMPTOW (1955), and DOUGLAS (1958) emphasize the influence of water discharge and turbidity on periphyton communities. An excellent review of the

artificial substrate technique was recently published



Fig. 2. View

Replicate slides were made for each measurement. The number of determinations, 5 for each ^{65}Zn analyses, 5 for ^{140}La total of 19 slides. The slides were stored awaiting instantaneous analyses. A fire in October before they could be scraped clean on the sampling area of 27.5 cm² drying to a constant weight after ignition at 525°C followed the method were filtered, adjusted to Beckman DU spectrophotometer determined with the corrected for unit area a mg dry weight/cm², & SLÁDEČKOVÁ (1964).

Several slides were

artificial substrate technique for investigating periphyton communities was recently published by SLÁDEČKOVÁ (1962).

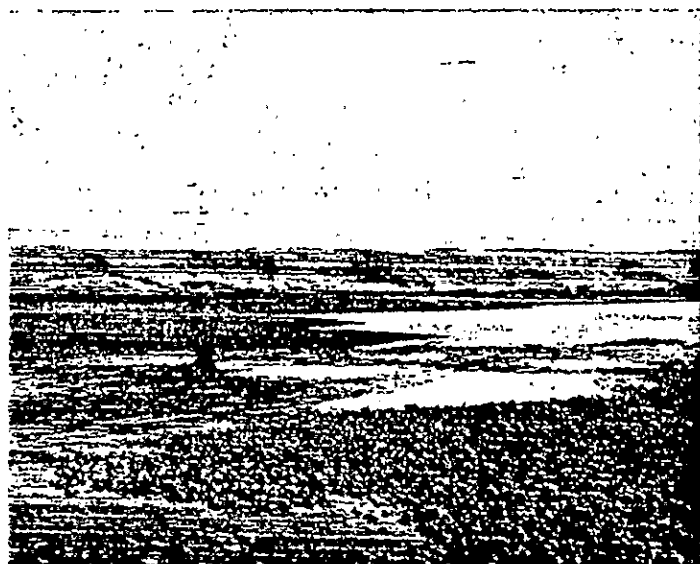


Fig. 2. View of study riffle at low water in fall.

Replicate slides were taken at each sampling for the various measurements. The normal allocation of slides was: 5 for gravimetric determinations, 5 for chlorophyll extractions, 3 combined for ^{32}P and ^{65}Zn analyses, 5 for ^{14}C productivity, and 1 for visual examination - a total of 19 slides. The series of slides used for ^{14}C productivity was stored awaiting installation of a Van-Slyke combustion train for analyses. A fire in our laboratory, unfortunately, destroyed them before they could be analyzed. The "frosted" end of each slide was scraped clean on both sides and discarded, leaving an effective sampling area of 27.5 cm^2 . Dry weights were determined by oven drying to a constant weight at 60° C . Ash-free dry weight was measured after ignition at 525° C . Chlorophyll extraction techniques generally followed the method of RICHARDS with THOMPSON (1952). Extracts were filtered, adjusted to 50 ml and absorbancies measured with a Beckman DU spectrophotometer. Chlorophyll concentrations were determined with the formulae of PARSONS & STRICKLAND (1963) and corrected for unit area. Net Production Rate (NPR) was calculated on a mg dry weight/ cm^2/day basis essentially as described by SLÁDEČEK & SLÁDEČKOVÁ (1964).

Several slides were pooled for radioactivity analyses; ^{65}Zn was

counted on a Packard Auto-Gamma single-channel spectrometer and ^{32}P by differential absorber techniques (PERKINS 1957). All samples were corrected for radioactive decay to the date of sampling; ^{32}P values were corrected for self-absorption.

An estimate of the amount of ^{32}P and ^{65}Zn transported by the river during each two-week exposure was calculated from analyses of integrated water samples. These were collected approximately 35 km downstream from the study area as part of a routine monitoring program, but were the only data of this type available. Data from the program are reported in curies per week and average picocuries per liter per week. Data for the correlations in this study were obtained by summing and averaging the appropriate figures. These data were available only for 1964. Solar radiation was measured with an Eppley pyrheliometer.

RESULTS AND DISCUSSION

Seasonal variations

Seasonal variations in the data are indicated in Table I. A summary of comparable measurements from other studies is given in Table II. Comparison of these values should be made with caution because of the different substrata used and the varying degree of community stabilization. The data from MCINTIRE et al. (1964) are presented as a comparison with studies of natural streams. The higher values found by McCONNELL & SIGLER (1959) can be partially attributed to the fact that they calculated their figures from measurements of algae on rocks within a square meter of stream bottom. These rocks would have a greater total surface area than one m^2 .

It is difficult to compare the radioactivity concentration data from this study with results of other workers since the exposure conditions and organisms sampled differ widely (BALL & HOOPER 1961; HARVEY 1963, 1965). DAVIS et al. (1958) reported a value of 66.0 nCi ^{32}P and 12.3 nCi ^{65}Zn per gram of wet *Stigeoclonium* in a single sample collected in August 1957 in the same area as the present study. DAVIS (1960) presents data showing the seasonal cycle of total beta emitters, in this case mostly ^{32}P , in sessile algae and plankton from the Columbia River. His data show periods of high radionuclide concentration in winter followed by low levels in spring. These changes are related to dilution by runoff. I have data (unpublished) on ^{32}P and ^{65}Zn concentrations in plankton from the Columbia showing essentially the same patterns. Data from the present study, however, differ slightly from these trends. Concentrations of ^{65}Zn show a similar pattern with high values during fall and early winter and low values

Analysis of periphyton and dissolved variables in the Columbia River, August 1963 to May 1964.

Sampling date	Dry weight mg/substrate	Ash weight mg/substrate	Ash-free dry weight mg/substrate	Chlorophyll <i>a</i> mg/substrate	Solar energy Langley's/2-wk. period	Net Production Rate mg dry wt/ cm^2/day	^{32}P nCi/substrate	^{32}P nCi/g dry wt.	^{65}Zn nCi/substrate	^{65}Zn nCi/g dry wt.	^{32}P River transport Ci/2-wk. period	^{65}Zn River transport Ci/2-wk. period	M.O. alkalinity ppm CaCO_3	pH	Water Temp C at time of sampling
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spectrometer and (1957). All samples were taken at the time of sampling; ³²P

ported by the river. From analyses of approximately 35 km routine monitoring data. Data from the ge picocuries per were obtained by These data were d with an Eppley

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eration data from exposure conditions PER 1961; HARVEY 66.0 nCi ³²P and a single sample sent study. DAVIS total beta emitters, from the Colum- ide concentration changes are related on ³²P and ⁶⁵Zn following essentially, however, differ n show a similar er and low values

Analyses of periphyton and limnological variables in the Columbia River, August 1963 to May 1964.

Sampling date	Dry weight mg/substrate	Ash weight mg/substrate	Ash-free dry weight	mg/substrate	Chlorophyll a mg/substrate	Solar energy Langley's/2-wk. period	Net Production Rate mg dry wt/cm ² /day	³² P nCi/substrate	³² P nCi/g dry wt.	⁶⁵ Zn nCi/substrate	⁶⁵ Zn nCi/g dry wt.	³² P River transport Ci/2-wk. period	⁶⁵ Zn river transport Ci/2-wk. period	M.O. alkalinity ppm CaCO ₃	pH	Water Temp C at time of sampling
8-5-63	19.89 ^a	12.17	7.72	—	—	8994	.050	—	—	.73	55.3	—	—	56	7.9	20
8-19	26.66	17.51	9.15	.13	.13	8158	.070	—	—	.81	31.1	—	—	58	7.9	22
9-3	27.45	18.85	8.60	.14	.14	7661	.065	—	—	1.62	54.7	—	—	56	8.1	23
9-16	8.99	5.42	3.57	.09	.09	5518	.025	—	—	.73	65.0	—	—	57	7.6	22.5
9-30	16.02	10.27	5.75	.10	.10	5860	.040	—	—	.69	40.7	—	—	58	7.9	21
10-14	8.33	5.44	2.89	.08	.08	4293	.020	—	—	.36	49.6	—	—	58	7.9	20.5
11-11	8.81	5.01	3.80	.08	.08	2426	.025	.39	46.5	.24	27.8	—	—	55	8.3	14.5
11-25	3.32	1.47	1.85	.04	.04	1404	.010	.40	106.9	.12	35.5	—	—	56	7.9	12
12-9	1.52	.24	1.28	.02	.02	1235	.005	.10	63.0	.06	47.0	—	—	57	7.5	13
12-23	3.51	2.86	.65	.02	.02	1253	.010	.09	55.4	.06	29.1	—	—	62	7.5	11.5
1-6-64	1.59	.50	1.09	.02	.02	1076	.005	.12	115.4	.08	48.7	180	462	58	7.6	10.5
1-20	1.78	.49	1.29	.01	.01	1510	.005	.06	58.2	.03	19.0	567	630	60	7.8	9
2-3	2.78	.83	1.95	.02	.02	2002	.005	.24	69.1	.04	19.5	357	553	60	7.5	9
2-18	11.37	4.62	6.75	.07	.07	2984	.030	1.39	96.5	.03	19.5	651	889	61	7.9	9
3-2	8.97	4.13	4.84	.04	.04	3700	.025	.67	68.9	.22	22.1	798	707	63	7.9	9.5
3-16	15.93	6.16	9.77	.09	.09	3634	.040	.97	55.6	.23	13.2	462	602	65	8.0	10
3-30	9.01	4.23	4.78	.04	.04	5091	.025	.88	110.6	.21	26.0	728	826	65	8.1	12.5
4-13	17.76	9.17	8.59	.05	.05	6117	.045	1.34	82.0	.50	30.5	840	1218	66	8.4	13
4-27	24.64	13.56	11.08	.09	.09	7024	.065	1.83	89.2	.68	33.1	546	700	67	8.3	15
5-11	10.43	6.44	3.99	.02	.02	7619	.025	.90	85.9	.41	41.0	1050	1190	66	8.2	11

a) Multiply substrate values by 363.6 to convert to sq. m.
b) ³²P extraction technique not perfected during first three months of study.

TABLE II

Mean values of biomass and chlorophyll *a* from the literature a.

Source	Locality and comments	Type of Substrate	Biomass (g dry wt/m ²)	Chlorophyll <i>a</i> (mg/m ²)
McCONNELL & SIGLER (1959)	Logan R., Utah	canyon section	25	300
		below canyon - fall		750
		below canyon - spring		1420
KOBAYASI (1961)	R. Arakawa, Japan	canyon section	2.5	30
		lower section	7.0	70
YOUNT (1956)	Silver Springs, Fla.	16 day exposure	Glass slides	34.7
WATERS (1961)	Valley Cr., Minn.	14 day exposure	Concrete cylinders	18.5
McINTIRE et al. (1964)	Artificial lab. streams	Developed at 6,000 lux	Rocks within measured area	.80
		Developed at 2,100 lux		.48
Present study		14 day exposure	Glass slides	4.2
				22.0

a) Data from McCONNELL & SIGLER and KOBAYASI calculated by them from chlorophyll-biomass ratios.

Data from YOUNT and WATERS calculated by me from data presented. The value from WATERS assumes that the periphyton uniformly covered the upper, illuminated half of the concrete cylinders.

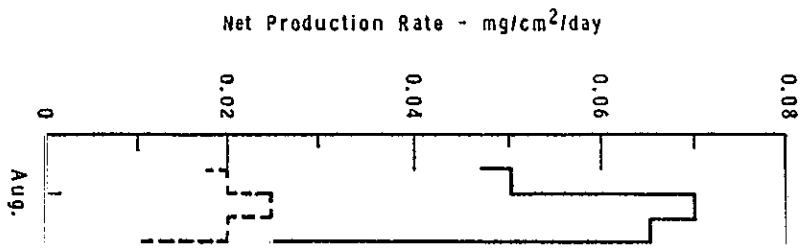


Fig. 3. Seasonal variation

The fall-winter periphyton community is mainly *Synedra* & *Cymbella*. The filamentous green algae becomes quite abundant through

in March, but a sharp apparent trend. The seasonal variation emphasized that these from a bare surface at community; they prob

in March, but ^{32}P concentrations fluctuated markedly with no apparent trend.

The seasonal variation of the NPR is shown in Fig. 3. It should be emphasized that these values represent a rate of production starting from a bare surface and not the two-week increment of an established community; they probably reflect maximal rather than 'normal' rates.

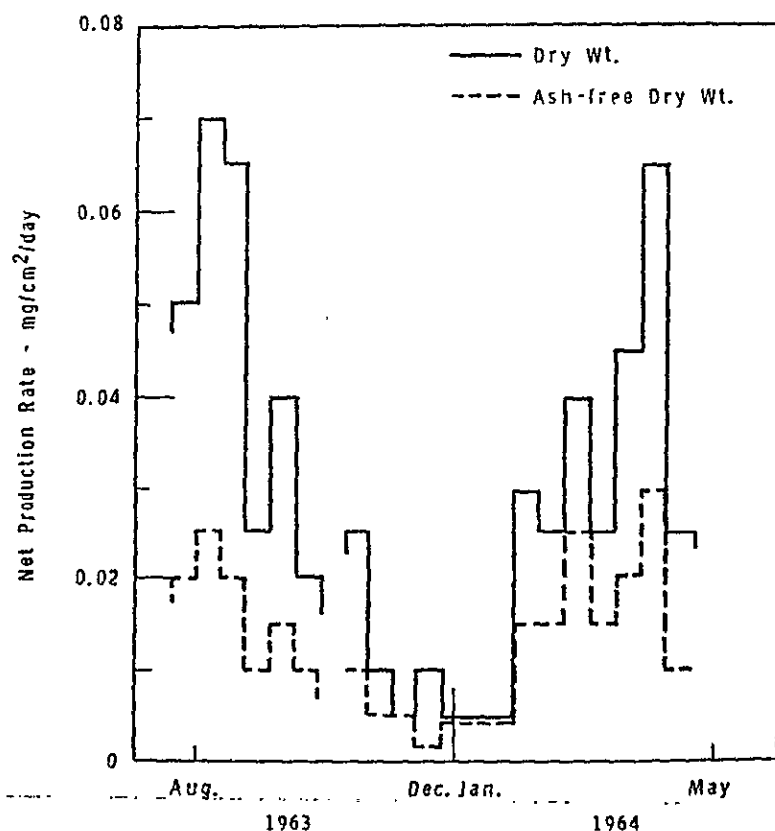


Fig. 3. Seasonal variation of Net Production Rate (NPR), August 1963 to May 1964.

The fall-winter periphyton community is dominated by diatoms - mainly *Synedra* and *Melosira*, but frequently with substantial populations of *Cymbella*, *Gomphonema*, *Fragilaria*, and *Achnanthes*. The filamentous green alga *Ulothrix zonata* (WEBER & MOHR) KUETZING becomes quite evident in late February and dominates the community through April. Diatoms again become dominant in May.

The significant components measured in this study of the ecological relationships (BLUM (1956, 1960)), interested reader is recommended to review their extensive influences on stream

[illegible]

Fig. 4. Correlation coefficients (r) between periphyton and limnological variables in the Columbia River, August 1963 to May 1964. Assuming a correlation coefficient equal to zero as a null hypothesis, all correlation coefficients are significantly different from zero at the 95 % level of confidence. NS = not significant. See Table I for units of measurement.

1. Biomass

Two commonly used estimates of the chlorophyll *a* content of a sample, the difference, - variously defined, in wet matter weight, or loss on drying, or on ash-free dry weight, or on ash-free ratio because of the difference in the community. The high ash: ash-free ratio is found when *Ulothrix* is dominant. A correlation coefficient of 0.925 (expressed as percent) was found by & BREHMER (1960) for the estimate of organic matter content. The chlorophyll *a* content is equally well with ash-free weight. KOBAYASHI (1960) found chlorophyll content of 1.0% in Arakawa.

2. Biomass, productivity

Solar radiation, measured at the water surface, had a correlation with the correlation between \ln when the many factors of community, non-composition, are correlated relatively high. Specific concentration and primary intensities and that rates at light saturation. SUGLER (1959) found production on con-

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Correlations

The significant correlation coefficients between the variables measured in this study are shown in Fig. 4. Excellent papers reviewing the ecological relationships of river algae have been published by BLUM (1956, 1960), BUTCHER (1932 et seq.) and others, and the interested reader is referred to these rather than taking the space here to review their extensive data concerning the effects of environmental influences on stream communities.

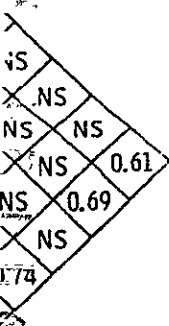
1. Biomass

Two commonly used indices of periphyton standing crop are: (1) the chlorophyll *a* content, and (2) gravimetric analyses – dry, ash, and the difference, – variously referred to as ash-free dry weight, organic matter weight, or loss on ignition. The low correlation between ash and ash-free dry weight is probably due to a lack of a constant ash: ash-free ratio because of major changes in the species composition of the community. The diatom-dominated community would have a high ash: ash-free ratio in fall and winter, but a lower value in spring when *Ulothrix* is dominant. SLÁDEČEK & SLÁDEČKOVÁ (1963) reported a correlation coefficient of .987 ($n = 33$) between air dry and oven dry weights, and .925 ($n = 34$) between dry and wet weight when expressed as percentage of mean values of lake periphyton. GRZENDA & BREHMER (1960) found that phytopigment units could be used to estimate organic matter weight of stream periphyton within certain limits. The chlorophyll *a* content in the present study correlated equally well with ash and dry weight, but less so with ash-free dry weight. KOBAYASI (1961) reported a linear relation ($n = 29$) between chlorophyll content and cell numbers of the periphyton in the River Arakawa.

2. Biomass, productivity, and environmental factors.

Solar radiation, measured as total Langley's (gram cal./cm²) incident at the water surface during the two week exposure, had a high correlation with the NPR and the chlorophyll *a* content. A better correlation between light and chlorophyll *a* might be expected, but when the many factors which influence chlorophyll content, e.g. age of community, non-functional chlorophyll, pre-nutrition, and species composition, are considered, the value found in this study might be relatively high. STEEMAN NIELSEN (1961) states that chlorophyll concentration and photosynthetic rate are directly related at low light intensities and that correlations between pigments and photosynthetic rates at light saturation are not a matter of course. McCONNELL & SIGLER (1959) found no close relationship between chlorophyll production on concrete 'rocks' placed in the Logan River, Utah, and

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insolation. The high correlation between NPR and chlorophyll *a* is consistent with the fact that closest relationships between pigment content and productivity are usually found only during periods of rapid growth. These conditions obtained in this study since colonization began from a bare surface and in two weeks time, the communities were probably still in the log-growth phase (unpublished data). RODHE, VOLLENWEIDER & NAUWERCK (1958) found a close relationship between net photosynthesis (^{14}C) and chlorophyll content during the spring phytoplankton increase but not in late summer. RYTHER & YENTSCH (1958) also found considerable variability between chlorophyll and net photosynthesis. WETZEL (1963) reports a poor correlation between productivity and estimates of mean periphyton biomass from pigment analyses.

Constant recording temperature instruments were unable to be placed in the riffle due to the extreme discharge fluctuations. The values used in the correlations were the average of the readings taken at the time a set of slides was placed in the river and when they were removed.

3. Radioactivity accumulation and environmental parameters.

Correlation coefficients between ^{32}P and ^{65}Zn accumulation and the biomass and environmental parameters are presented using two measurements: (1) radioactivity per substrate, and (2) radioactivity per gram dry weight. Significant correlations were obtained only when using the radioactivity per substrate values (Fig. 4). The amount of ^{32}P and ^{65}Zn accumulated was highly correlated with dry and ash weight; ^{65}Zn accumulation had a slightly higher correlation with chlorophyll *a* content than did ^{32}P . The low correlation between solar radiation and radionuclide accumulation was probably due in part to its indirect influence through chlorophyll and biomass production.

Very little appears in the literature concerning radionuclide accumulation by periphyton (except see BALL & HOOPER 1961); however, some work has been done with other algae, mostly marine. GUTKNECHT (1961, 1963, and 1965) observed that light and temperature effects and ^{65}Zn uptake are related to metabolic accumulation by photosynthesis; that low temperatures retard ^{65}Zn uptake in living and killed *Ulva*; and that the relationship between ^{65}Zn uptake and photosynthesis in seaweeds may be indirectly related to pH changes. BACHMANN & ODUM (1960) found that ^{65}Zn was taken up in proportion to the gross oxygen production and accumulated in proportion to net oxygen production in marine seaweeds.

GEST & KAMEN (1948) observed no strict proportionality between phosphate uptake and the over-all metabolic level of *Chlorella* and *Scenedesmus*. WHITFORD and SCHUMACHER (1964) found that *Spiro-*

gyra and *Oedogonium* to speeds up to 40 cm/sec.

One other factor which isotope to which the periphyton showed that the concentration of the Columbia River at activity in the water than

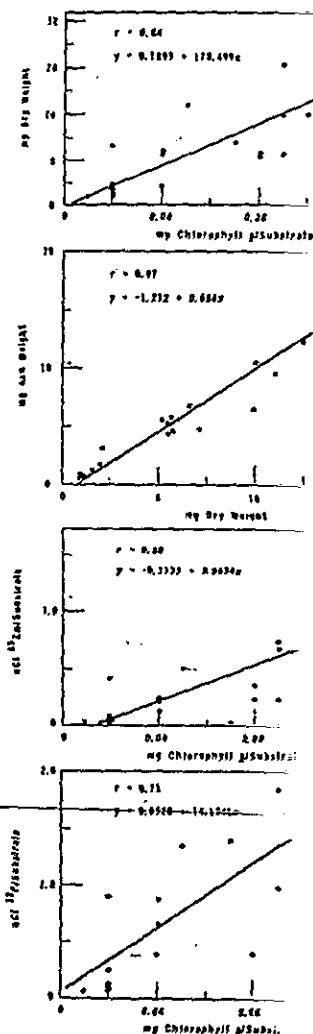


Fig. 5. Scatter diagrams a.

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One other factor which must be considered is the amount of each isotope to which the periphyton was exposed. DAVIS et al. (1956) showed that the concentration of radioisotopes in the phytoplankton of the Columbia River at Hanford was more closely related to radioactivity in the water than to certain environmental features. Thus, it

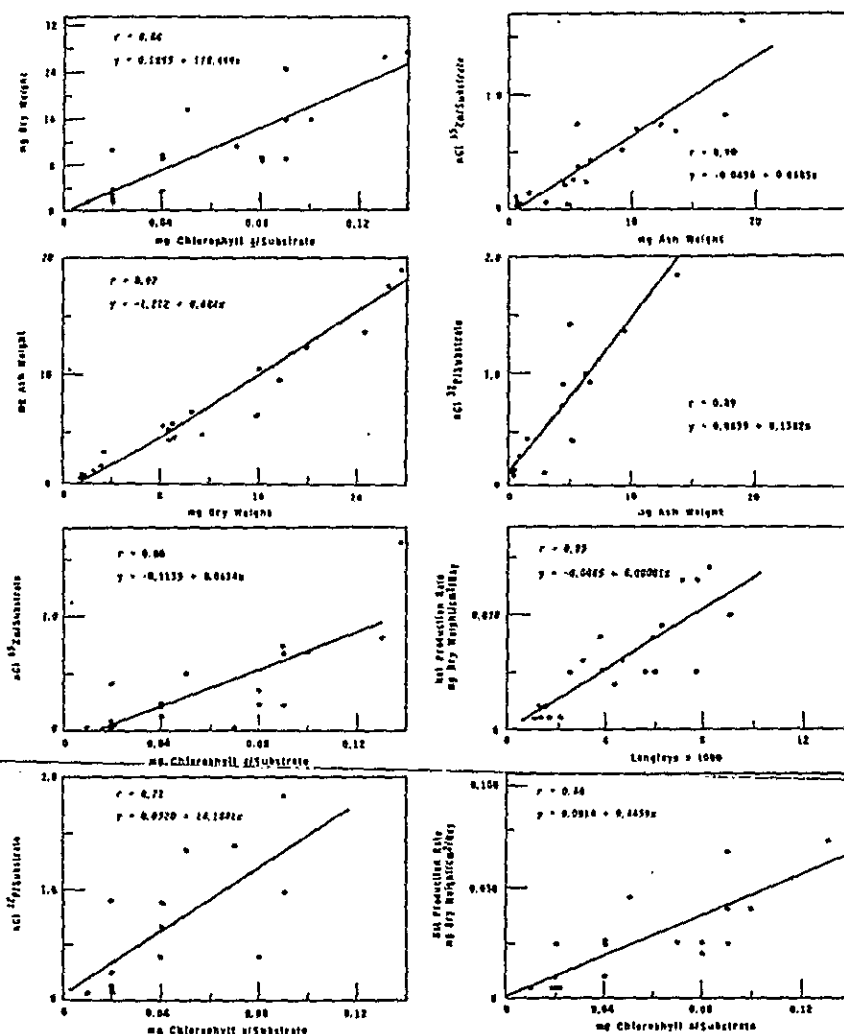


Fig. 5. Scatter diagrams and regression lines of selected data, August 1963 to May 1964.

was suspected that the river radioisotope burden might influence the uptake by the periphyton to such a degree as to mask out sorption patterns related to biological activity. In the present study, surprisingly, there were no significant correlations found between the amount of ^{32}P and ^{65}Zn per substrate and the corresponding activity in the water whether it was expressed as total curies transported or average pCi/l during the two week exposure. The river burden data, since they were not taken as a part of this study and included particulate as well as dissolved matter, may explain the low correlations.

Representative scatter diagrams and regression lines from the data are illustrated in Fig. 5 and suggest that the relationships observed were essentially linear under the conditions studied.

Mode of uptake

The fact that the radionuclide accumulation had a higher correlation with dry and ash weight than to ash-free dry weight lends support to the evidence that the dominant mode of uptake is adsorption rather than absorption (DAVIS et al. 1958; ODUM et al. 1958; BACHMANN 1961; GUTKNECHT 1965). A higher correlation with the organic fraction would be expected if absorption were the prevalent method of uptake.

ACKNOWLEDGMENTS

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SUMMARY

An investigation of the periphyton of the Columbia River below the Hanford Atomic Works, Washington, was conducted to study the relationships between productivity, radionuclide uptake, and environmental influences.

Best correlations between the four biomass measurements were between dry weight, ash weight, and chlorophyll *a*. Net Production Rate varied from 0.005 to 0.070 mg dry weight/cm²/day and was closely related to chlorophyll *a* and also to solar energy.

The accumulation of ^{32}P and ^{65}Zn was highly related to dry and ash weight and chlorophyll *a*. Low correlations were found between radionuclide accumulation and the radioisotope burden of the river. The data suggest that adsorption was the dominant mode of uptake.

Une investigation des periphytes de la Columbia River, sous les Hanford Atomic Works, Washington, a été conduite pour étudier les rapports entre la productivité, l'adsorption des radionucléides et les influences des environnements.

On a réalisé les meilleures corrélations entre les quatre mesures de biomasse de la masse biologique pour le poids sec, le poids des cendres et la chlorophylle *a*. Le Taux de Production variait de 0.005 à 0.070 mg poids sec/cm²/jour et variait de 0.005 à 0.070 mg poids sec/cm²/jour et était étroitement lié à la chlorophylle *a* et aussi à l'énergie solaire.

Le rapport meilleur pour l'adsorption des radionucléides était le poids sec et le poids des cendres. Les relations petites entre le radionucléide accumulé et le poids sec et le poids des cendres au sein du fleuve des radionucléides suggèrent que l'adsorption était la mode dominante d'adsorption.

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RÉSUMÉ

Une investigation des periphytons du Columbia River en bas de Hanford Atomic Works, Washington, a été faite dans le but d'étudier les rapports entre la productivité, l'incorporation des radioéléments et les influences des environs.

On a réalisé les meilleurs des correlations entre les quatre mesures de masse biologique pour le poids sec, le poids de cendres, et la chlorophylle *a*. Le Taux du Production Net (Net Production Rate) a varié de 0.005 à 0.070 mg du poids sec/cm²/jour, montrant une dépendance proche de la chlorophylle *a* et également de l'énergie du soleil.

Le rapport meilleur pour le dépôt du ³²P et du ⁶⁵Zn était à le poids sec et le poids de cendres, et à la chlorophylle *a*. On a trouvé des correlations petites entre le dépôt des radioéléments et la concentration au sein du fleuve des radioéléments. Les données impliquent que l'adsorption était la mode prédominante pour l'incorporation.

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